Interferon-beta therapy increases concentration of soluble Interferon alpha/beta receptor in Multiple Sclerosis patient serum.

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Abstract:

Interferon-beta is a frontline treatment for Multiple Sclerosis (MS) and yet the molecule's mechanism of action in this disease is not fully understood. We have examined the serum/plasma concentrations of soluble Interferon alpha/beta receptor 2 (IFNAR2a) using a newly developed IFNAR2a ELISA in 35 normal human donors, 25 MS patients not on IFN therapy, and 29 MS patients on IFN therapy. Other biomarkers, IFN-beta, and Type I IFN activity were also assessed in most samples. In most patients receiving IFN-beta therapy, IFN beta mass as well as IFN bioactivity can be detected in the serum. Consistent with previous literature, IFN bioactivity is lower than that which would be predicted based on IFN beta mass. For patients on IFN-beta therapy, we observe that IFNAR2a concentration is significantly ($p<0.0001$) higher than that in MS patients on IFN-free therapy and relative to normal donors ($p=0.0002$). Normal donor IFNAR2a concentration does not significantly differ from that of MS patients on IFN-free therapy. These results expand upon previous literature which suggests that the mRNA encoding a soluble form of the IFN receptor is increased by long term IFN-beta therapy. Data from additional samples, potential mechanisms, and correlations to other biomarkers will be discussed.

Introduction

Interferons (IFN) have been identified as important immuno-modulators in autoimmune diseases. IFN-beta for one, is the most accepted bio-therapeutic for the treatment of multiple sclerosis (MS) and has shown to decrease relapses, brain lesions, and slow neuro-degeneration in patients [1]. However, the clinical response to IFN-beta is highly variable [2]. Hence, understanding the mechanism of action of IFN-beta in MS treatment may prove to be highly valuable in improving the efficacy of this therapy.

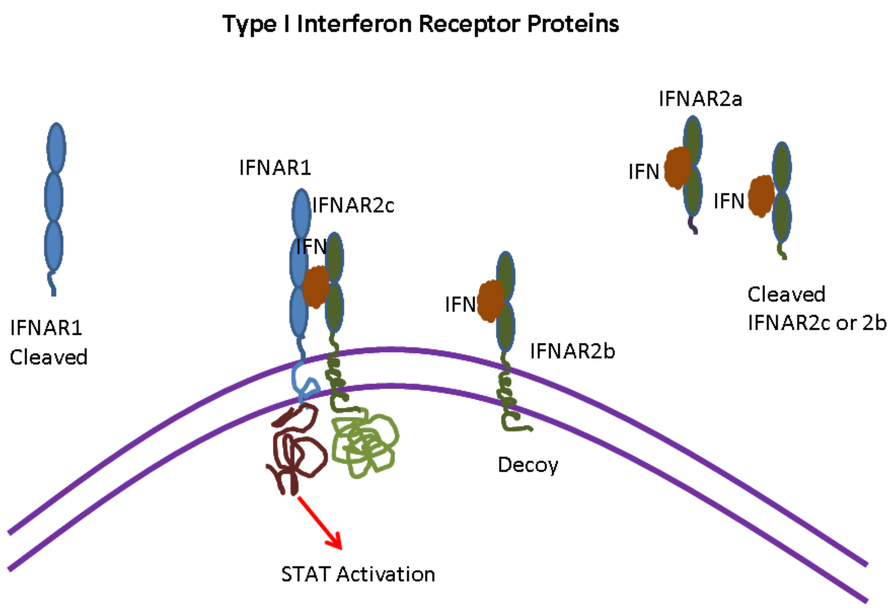
IFN-beta activates various signaling cascades via its high affinity interaction with the multi-subunit type-I IFN cellular membrane receptor (IFNAR). The IFNAR2 subunit exists in 2 transmembrane isoforms (IFNAR2b and IFNAR2c) and a soluble form (IFNAR2a) arising from the alternative splicing of the mRNA and/or post-translational proteolytic cleavage of the transmembrane isoforms [3]. IFNAR2a may act as an antagonist or serve to stabilize IFN-beta by the formation of a complex *in vivo* [4].

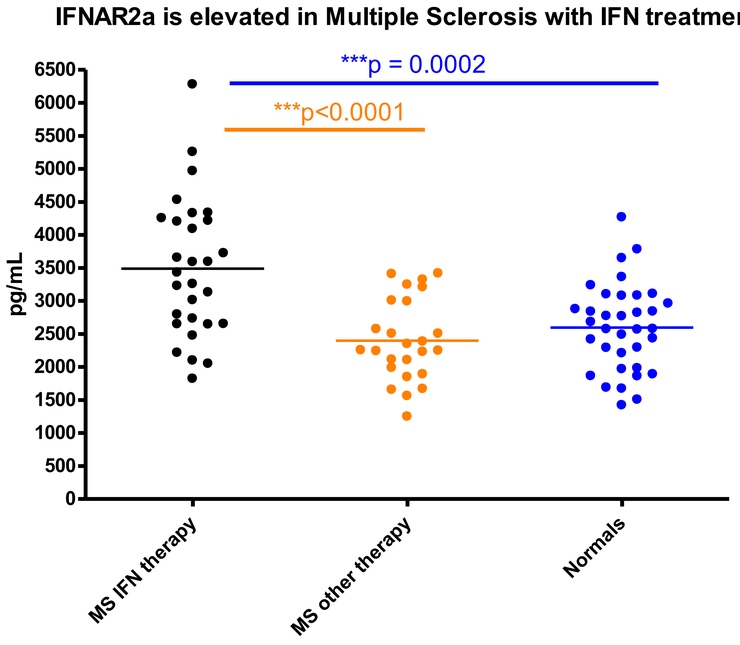
We measured the concentrations of IFNAR2a and IFN-beta in the sera/plasma of 35 normal human donors, 25 MS patients not on IFN therapy, and 29 MS patients on IFN therapy (Avonex, Rebif, or Betaseron). Type-I IFN activity in the MS samples was also measured using a cell based assay. A panel of other cytokines were also examined using a 16-plex and a 9-plex ELISA assay.

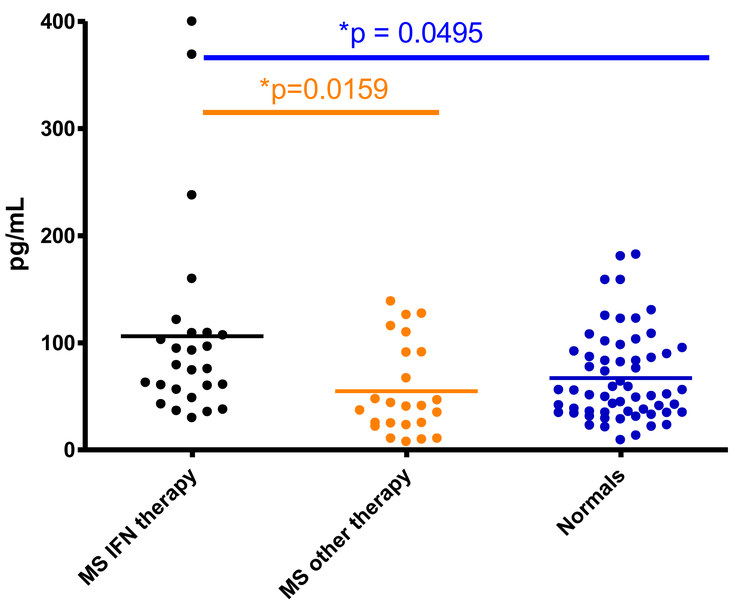
Particularly, the data from this study suggests that prolonged treatment with IFN leads to an elevated concentration of IFNAR2a in MS patient sera.

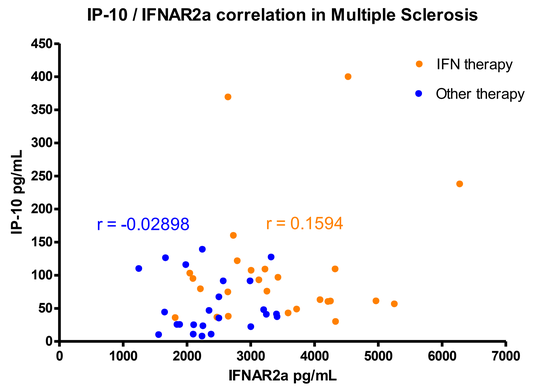
Multiple Sclerosis Serum/Plasma Results

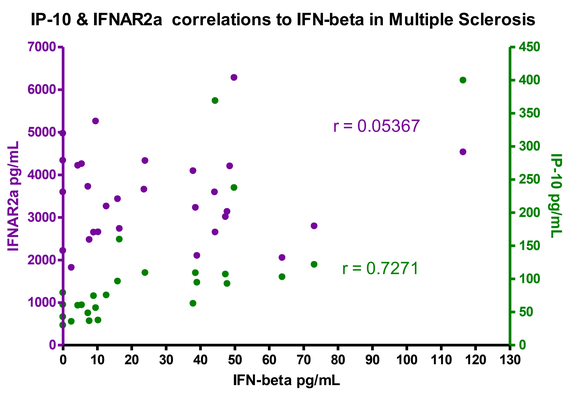
Lot#	IFN Treatment	sIFNAR2 pg/mL	IP-10 pg/mL	IFN-beta pg/mL
6260	None	1249.47	109.75	0.00
6261	None	1986.95	115.56	7.08
6262	None	1654.62	43.81	0.00
6263	None	2256.28	23.21	0.00
6264	Rebif	3012.31	106.88	47.33
6265	Rebif	2653.87	37.49	10.25
6266	None	2104.28	10.44	0.00
6267	None	1891.80	25.22	0.00
6268	None	2240.64	7.52	0.00
6269	None	1848.72	25.06	0.00
6270	None	2507.16	34.80	0.00
6271	None	2350.60	46.36	0.00
6272	Avonex	4256.12	60.48	5.51
6273	Avonex	2645.71	74.29	8.95
6274	None	2573.63	90.87	0.00
6275	None	1561.73	9.81	0.00
6276	Avonex	4532.48	399.63	116.46
6277	None	3249.66	40.38	0.00
6278	Rebif	4329.03	109.04	23.95
6279	Avonex	5257.40	56.28	9.58
6280	None	3418.63	36.75	0.00
6281	Betaseron	3724.02	48.32	7.33
6282	Avonex	3227.97	108.85	38.60
6283	Rebif	6279.80	237.55	49.82
6284	Rebif	4091.49	62.64	37.92
6285	None	3208.44	47.39	0.00
6286	Rebif	2648.96	368.91	44.33
6287	None	2111.83	24.77	0.00
6288	Betaseron	3432.08	96.29	15.93
6289	None	3324.19	127.14	0.00
6290	Rebif	2049.60	102.78	63.80
6291	None	2384.66	10.57	0.00
6292	Avonex	4335.72	29.73	0.00
6293	Rebif	4215.79	59.76	4.36
6317	None	3006.28	21.65	0.00
6318	Avonex	2794.67	121.42	73.13
6319	Avonex	4201.80	na	48.56
6320	None	2229.61	na	0.00
6321	None	2995.94	91.03	0.00
6322	Avonex	3592.03	42.53	0.00
6323	Rebif	2215.53	79.00	0.00
6324	Avonex	1821.77	35.42	2.49
6325	Avonex	2476.70	36.29	7.72
6326	None	3408.39	40.93	0.00
6327	Avonex	3132.53	92.60	47.81
6328	None	2247.19	138.67	0.00
6329	Avonex	2099.33	94.47	39.02
6330	Rebif	3259.64	75.37	12.65
6331	Betaseron	3656.96	na	23.63
6332	Avonex	4969.10	60.82	0.00
6333	Avonex	2733.21	159.67	16.47
6334	None	1668.82	125.98	0.00
6335	None	2504.93	66.89	0.00
6336	Rebif	3591.84	na	44.14

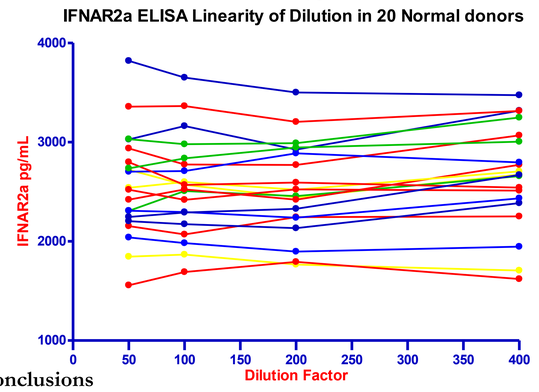












Conclusions

Interferons exhibit a wide a range of biological activities including antiviral, anti-proliferative, and other immunomodulatory effects. In the treatment of multiple sclerosis, IFN-beta is thought to play an anti-inflammatory role by regulating the movement of different immune cells across the blood brain barrier as well as promoting the production of other anti-inflammatory factors [5]. However, the exact mechanism of action is more involved and is still unclear. In this study, we observed that the concentration of the soluble version of the type-I interferon receptor was elevated in the serum samples from MS patients that received interferon therapy. This could be a response to neutralize excess interferon activity from prolonged interferon treatment because cellular receptors are known to undergo modification post-ligand binding to limit cell response. However, transcriptional regulation may also be one of the sources of the soluble receptor after IFN stimulation. An increase in the IFNAR2a mRNA in MS patients on long term IFN-beta therapy has been reported [7].

We were able to measure IFN-beta (mass) in 86% of the samples from IFN treated MS patients and noted that IFN-beta concentration in the samples did not correlate well to the IFNAR2a levels. This suggests a more indirect induction of soluble receptor expression from IFN treatment or that the half-lives of the two analytes are vastly different. Nonetheless, IFNAR2a levels may be indicative to the extent of progress/failure of IFN therapy in multiple sclerosis.

We also observed that IP-10 (or CXCL10) is elevated in MS serum from patients that received IFN treatment compared to patients with other therapies. Moreover, a statistically significant correlation (bivariate pearson) was observed between concentrations of IFN-beta and IP-10 in these samples. This confirms previously reported observation that IFN-beta administration in MS patients leads to a transient increase in certain chemokines including IP-10. Although IP-10 is pro-inflammatory chemokine that supports monocyte/neutrophil infiltration, the IFN-beta induced transient systemic upregulation may alter the local concentration gradients of this chemokine in tissues to produce an anti-inflammatory effect [6].

References

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